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# Evidence for a Programmed Circannual Life Cycle Modulated by Increasing Daylengths in *Neanthes limnicola* (Polychaeta: Nereidae) From Central California

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**Abstract.** Timing of parturition, fecundity, and life span were determined in laboratory cultures of the semelparous, self-fertilizing, viviparous polychaete *Neanthes limnicola*. Worms were exposed to fixed daylengths (short—8h light: 16h dark; neutral—12h:12h; long—16h:8h), switched between different fixed daylengths, and switched from fixed daylengths to increasing or decreasing daylengths. Timing of parturition was synchronized when under neutral daylength, but became asynchronous under both short and long daylength, as well as when any of the fixed daylength was followed by decreasing daylengths. Worms under neutral daylength had the highest fecundities and shortest life spans, while those under long days had the lowest fecundities and longer life spans. When fixed daylength (short, neutral, long) was followed by increasing daylengths, timing of parturition was synchronized, fecundity was high, and life span shortened. These and earlier published experiments on the influence of seasonally changing photoperiods indicate that the life cycle of the estuarine *N. limnicola* is programmed to be completed in somewhat less than a year, and that seasonally changing photoperiods modulate it to determine the optimal time of parturition.

## Introduction

Photoperiodic control of reproduction has been demonstrated for many organisms (reviewed by Saunders, 1982; Gwinner, 1986). Most experimental work has focussed on the effects of fixed daylength on annual reproductive rhythms (*e.g.*, for annelids: Garwood and Olive, 1982; Olive and Pillai, 1983; Clark, 1988; Schierwater and Hauenschild, 1990). However, seasonally changing photoperiod also has profound effects on the timing of reproduction (Goss, 1982, 1984; Pearse *et al.*, 1986). Such is the case for *Neanthes limnicola* (Johnson, 1901), a viviparous, self-fertilizing hermaphroditic polychaete that gives birth mainly during the spring (late February–May) in the brackish-water creeks flowing into Monterey Bay, California (Smith, 1950; Fong and Pearse, 1992). This semelparous worm responds to seasonally changing photoperiod by giving birth to young in spring light regimes when maintained under either in-phase or 6 months out-of-phase light conditions. Worms in culture live for 6 months to 2 years and still reproduce mainly in spring light regimes (Fong and Pearse, 1992). These findings suggested to us that the worms must "see" either one or more critical daylengths, or increasing daylengths mimicking spring light regimes, to complete sexual maturation. The present paper reports on experiments that examined the effects of constant fixed daylength, and fixed daylengths followed by increasing or decreasing daylengths, on the timing of parturition, on fecundity, and on life span in culture of *N. limnicola*. These experiments revealed evidence for an endogenous, circannual rhythm that responds to increasing daylengths.

## Materials and Methods

We have maintained worms and their offspring in the laboratory since October 1987 when approximately 20 adults were collected from Watsonville Slough, California (36°45′N; 121°45′W). Worms used in all experiments

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were born in the laboratory as a result of self-fertilizations from lab-reared adults. Although worms in the field are usually born in the spring, birth date in the laboratory can be controlled by manipulating seasonally changing daylengths (Fong and Pearse, 1992). Juvenile worms were reared singly, initially in small, plastic petri dishes, then transferred to 80 × 100 mm pyrex culture dishes with lids (1 worm/dish). All culture dishes were maintained at laboratory air temperature (Fig. 1), and a salinity of 15‰. Worms were fed brine shrimps once per week, and the culture media was changed 1–2 days after each feeding. As worms grew and matured they were monitored daily for signs of reproduction. About 10 days before giving birth, the body wall of the adult becomes greenish and semi-transparent, and developing embryos are easily seen moving through the coelomic fluid. At birth, juvenile worms emerge through fissures in the degenerating body wall of the dying adult. Occasionally, adult worms with normal body size and reproductive morphology produced no young. Because they looked and behaved normally before parturition, these worms (n = 7) were included in all statistical analyses. Parturition date, life span (days adults spent in culture) and fecundity (numbers of young produced) were recorded for each birth. All experiments (treatments and numbers of worms used) are summarized in Table I. For statistical analysis of parturition dates, sequential numerical values were assigned to each date in each experiment; the value of 1 was assigned to the date of the first parturition, 2 to the following calendar day, etc. (*e.g.,* in experiment A the first parturition occurred on 2 April 1989 and was given the value 1, 3 April = 2, and 5 May = 33, etc.). Analysis of variance was used to compare mean parturition dates, life span, and fecundity (see Fong, 1991, for full analysis).

*Fixed daylength (experiment A)*

Immediately after birth (Aug–Sept 1988), each of the 108 worms was placed in one of three light-tight wooden boxes (n = 36/box) illuminated with fluorescent lights (General Electric F40 daylight) at fixed photoperiods of either long (L:D 16h:8h), neutral (12h:12h), or short (8h:16h) daylengths. The northern distribution of *N. limnicola* extends to Vancouver Island, where extreme daylengths of 16h:8h and 8h:16h occur in June and December, respectively. In central California, extreme daylengths are approximately 14.5h:9.5h and 9.5h:14.5h. After 3 months, some of the worms in each light treatment were removed from their boxes and switched into one of the other two light regimes (for example, 24 worms were removed from the neutral daylength box, and 12 each were placed under short and long daylengths; symbolized by neutral → short and neutral → long). The remainder of the worms were maintained under their initial fixed-daylength light regimes. Of the 108 worms, 27 (25%) died during the experiments either before or after shifting light regimes (Table I).

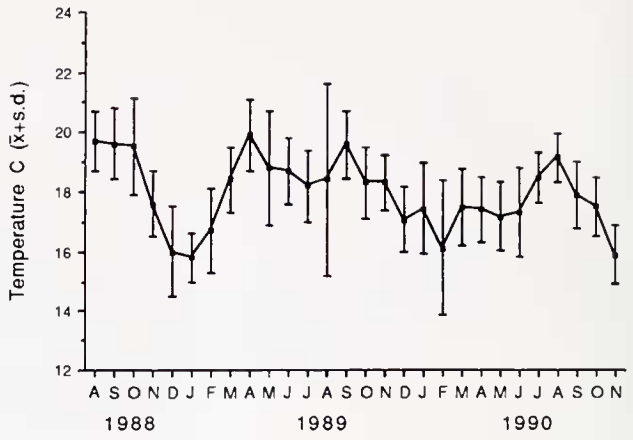


**Figure 1.** Monthly air temperature ($\bar{x}$ + S.D.) at Long Marine Laboratory from daily records.

*Fixed daylength followed by increasing daylengths (experiment B) or by decreasing daylengths (experiment C)*

In experiment B, 60 worms (all born on 4 May 1989) were divided equally among 3 fixed daylength light regimes of long (L:D 16:8), neutral (12:12), or short (8:16) daylength. After 3 months, roughly half of the worms in each light treatment were switched into a room where daylengths would be increasing for almost 5 months, (light regimes corresponding to February–June), controlled by a mechanical warehouse clock switch (Astronomic Time Switch, R.W. Cramer & Co., Type SY Model SOL). The lights were turned on and off at local sunrise and sunset 6 months out of phase with ambient photoperiod. The early February light regime was approximately (L:D) 10:14, thus worms initially exposed to neutral daylength (12:12) saw a shorter, but increasing daylength for the first 6 weeks after shifting. Those initially exposed to long (16:8) daylength also saw shorter, but increasing daylengths. Only worms initially exposed to short (8:16) daylength saw longer and increasing daylengths. The other half of the worms in each treatment were maintained in their original fixed-daylength light regimes (control). Of the 60 worms, 17 (28%) died either before or after shifting (Table I).

In experiment C, 60 worms (born 30 July–5 August 1989) were kept in phase (decreasing daylength) for about 1 month after birth, then divided equally and maintained as above in either short, neutral, or long daylengths for 7 weeks. In late September, 10 worms from each treatment were transferred to a room where daylengths (controlled by another time switch) would be decreasing (in phase)

**Table I**

*Mean parturition dates, mean numbers of young, and mean days in culture of* Neanthes limnicola *in all experiments*

| Experiment name | Photoperiod treatment | Date adults born | n surviving | Mean parturition date | Mean # young (+S.D.) | Mean life span (+S.D.) |
|---|---|---|---|---|---|---|
| A. Fixed daylength | short | 22 Aug 88 | 6 | 4 May | 100.6 (60.4) | 252.6 (18.0) |
| | neutral | 22–28 Aug 88 | 8 | 12 July | 163.8 (44.3) | 318.5 (141.8) |
| | long | 22 Aug 88 | 10 | 11 July | 114.4 (44.5) | 313.3 (38.5) |
| | short → neutral | 22 Aug 88 | 7 | 3 June | 124.4 (41.0) | 281.4 (92.3) |
| | short → long | 22 Aug 88 | 7 | 10 June | 124.1 (49.5) | 286.4 (59.8) |
| | neutral → short | 28 Aug 88 | 11 | 11 May | 153.7 (25.7) | 254.2 (15.1) |
| | neutral → long | 28 Aug, 14 Sept 88 | 12 | 10 Aug | 99.91 (56.1) | 340.2 (97.1) |
| | long → short | 22 Aug 88 | 9 | 11 July | 139.7 (55.8) | 309.6 (55.8) |
| | long → neutral | 22 Aug 88 | 11 | 15 Aug | 92.3 (77.4) | 349.9 (102.6) |
| | Independent of shifting | | | | | |
| | initially short | | 20 | 28 May | 117.2 (48.8) | 274.4 (64.2) |
| | initially neutral | | 31 | 1 July | 135.5 (51.7) | 304.0 (98.6) |
| | initially long | | 30 | 24 July | 113.9 (60.0) | 325.6 (72.8) |
| | Pooled | | | | | |
| | pooled short + neutral | | 51 | 18 June | 128.3 (50.9) | 292.4 (87.3) |
| | pooled long | | 30 | 24 July | 113.9 (60.0) | 325.6 (72.8) |
| B. Fixed → increasing daylength | short | 4 May 89 | 6 | 1 Apr | 87.2 (68.7) | 332.6 (141.5) |
| | neutral | 4 May 89 | 7 | 1 Apr | 143.4 (66.0) | 330.3 (18.7) |
| | long | 4 May 89 | 4 | 23 May | 64.6 (56.9) | 385.0 (113.0) |
| | short → inc | 4 May 89 | 7 | 29 Nov (=May light regime) | 105.8 (48.5) | 209.4 (21.5) |
| | neutral → inc | 4 May 89 | 10 | 14 Dec (=June light regime) | 115.5 (35.8) | 223.3 (8.1) |
| | long → inc | 4 May 89 | 9 | 12 Jan (=July light regime) | 152.8 (46.5) | 253.1 (7.4) |
| C. Fixed → decreasing daylength | short | 5 Aug 89 | 9 | 22 July | 89.22 (55.9) | 350.7 (116.1) |
| | neutral | 5 Aug 89 | 9 | 27 April | 108.9 (36.7) | 265.0 (5.1) |
| | long | 30 July 89 | 5 | 14 Aug | 39.4 (41.2) | 380.4 (32.7) |
| | short → dec | 5 Aug 89 | 10 | 20 Aug | 89.2 (40.4) | 377.3 (131.8) |
| | neutral → dec | 5 Aug 89 | 8 | 20 May | 106.3 (18.5) | 288.3 (25.4) |
| | long → dec | 30 July 89 | 8 | 22 June | 77.87 (30.2) | 327.3 (37.6) |
| | Independent of shifting | | | | | |
| | initially short | | 19 | 6 Aug | 89.2 (46.9) | 364.7 (121.9) |
| | initially neutral | | 17 | 8 May | 107.6 (28.7) | 275.9 (21.0) |
| | initially long | | 13 | 13 July | 63.1 (38.4) | 347.7 (43.7) |

Parturition dates are dates on which adults gave birth. Daylengths are: short daylength = (L:D 8h:16h), neutral daylength = (12h:12h), long daylength = (L:D 16h:8h). Short → neutral indicates cultures which were initiated in (8:16), then after 3 months shifted into 12:12, etc. inc = increasing daylengths; dec = decreasing daylengths.

for about 3 months. On 23 December 1989, worms were placed in constant short daylengths (8:16) where they remained until they gave birth. Control worms (n = 10 for each light treatment) were maintained in their original fixed-daylength light regimes. Of the 60 worms, 11 (18%) died during the experiment (Table I).

## Results

### *Fixed daylength (experiment A)*

Photoperiod significantly affected the timing of parturition in *Neanthes limnicola* (one-way ANOVA of mean parturition date, $F_{8,72} = 2.08$, $P = 0.04$). Worms maintained in constant short and neutral → short daylengths gave birth on average in the ambient spring (May), and those in short → neutral and short → long in the ambient late spring (June) (Table I, Fig. 2). Worms maintained in constant neutral (12:12) also gave birth in the ambient spring, but parturition dates were separated by 10–11 months (Fig. 2); thus the mean parturition date was in July. Those worms that encountered long daylengths initially, gave birth in July and August on average (Table I, Fig. 2). Four significant comparisons were made between groups (Fisher's Least Significant Difference test, $P < 0.05$), and all involved groups that saw either short or neutral daylengths initially, compared with those that saw long daylengths at any stage. In each case, exposure to long daylength resulted in worms giving birth significantly

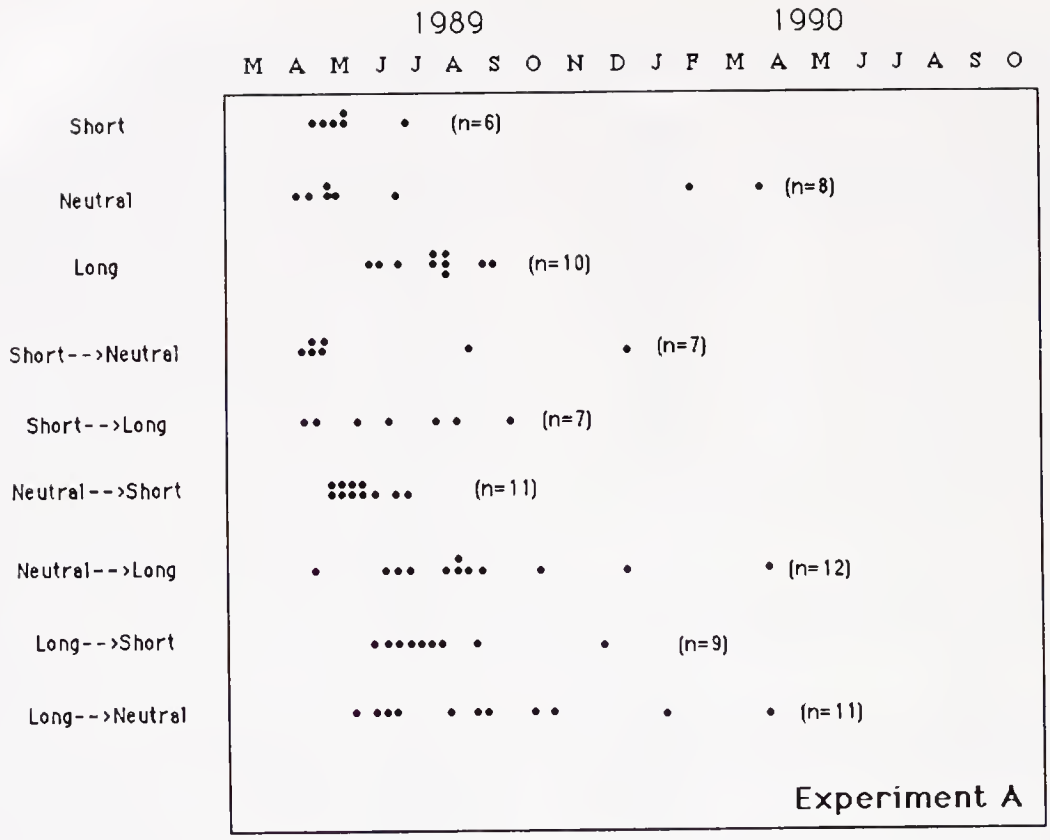


**Figure 2.** Parturition dates of *Neanthes limnicola* in various conditions of fixed daylength (experiment A). Each point represents a parturition. Daylengths are short = (L:D 8:16), neutral = (L:D 12:12), and long = (L:D 16:8). Short → neutral indicates cultures initiated in short daylength, then shifted into neutral daylength, etc. Adults that gave birth were themselves born in August–September 1988.

later in the calendar year, and spread over an extended period. Despite significant differences in the timing of parturition, photoperiod did not significantly affect life span (one-way ANOVA, $F_{8,72} = 1.75$ $P = 0.10$), but did have a significant effect on fecundity (one-way ANOVA, $F_{8,72} = 2.20$ $P = 0.04$). Highest fecundities were recorded in worms that saw either constant neutral ($\bar{x} = 163.8$, n = 8) or neutral → short ($\bar{x} = 153.7$, n = 11) daylengths (Table I). Lowest fecundity was in the long → neutral treatment ($\bar{x} = 92.27$, n = 11); 4 of these 11 worms had good reproductive morphology, but produced no young.

The initial daylengths to which worms were exposed (*i.e.*, independent of shifting) had a significant effect on timing of parturition (one-way ANOVA of mean parturition date, $F_{2,78} = 3.03$, $P = 0.05$); worms in short and neutral daylengths gave birth in the spring and early summer ($\bar{x}_{short}$ = 28 May, $\bar{x}_{neutral}$ = 1 July, Table I), but worms that initially saw long daylengths gave birth in the midsummer ($\bar{x}_{long}$ = 24 July; Fisher's LSD, $P < 0.05$ for both comparisons). However, no significant difference in life span or fecundity exists among these photoperiodic groups.

*Increasing daylength experiments (experiment B)*

The timing of parturition was affected by increasing daylength (one-way ANOVA of mean parturition date, $F_{5,37} = 7.79$, $P = 0.0001$). Worms that saw increasing daylength gave birth in late spring-early summer light regimes ($\bar{x}_{short\rightarrow inc}$ = 29 November = May light regime, $\bar{x}_{neutral\rightarrow inc}$ = 14 December = June light regime, $\bar{x}_{long\rightarrow inc}$ = 12 January = July light regime; Table I), and in each treatment, most of the parturition dates were clustered within 1–2 months of each other (Fig. 3). Worms in constant neutral daylength showed a trend similar to those that saw increasing daylengths, giving birth in the ambient spring ($\bar{x}$ = 1 April) with all births clustered within 2 months of each other (Fig. 3). Most worms in constant short daylength gave birth from October 89 to May 1990 ($\bar{x}$ = 1 April); one worm lived in culture until November 1990, had good reproductive morphology, but produced no young. Of the four worms in constant long daylength, three gave birth from June to September 1990 ($\bar{x}$ = 22 Aug.); one worm lived until late December 1989 but produced no young. In the latter two light treatments, no consistent pattern of parturition timing was evident.

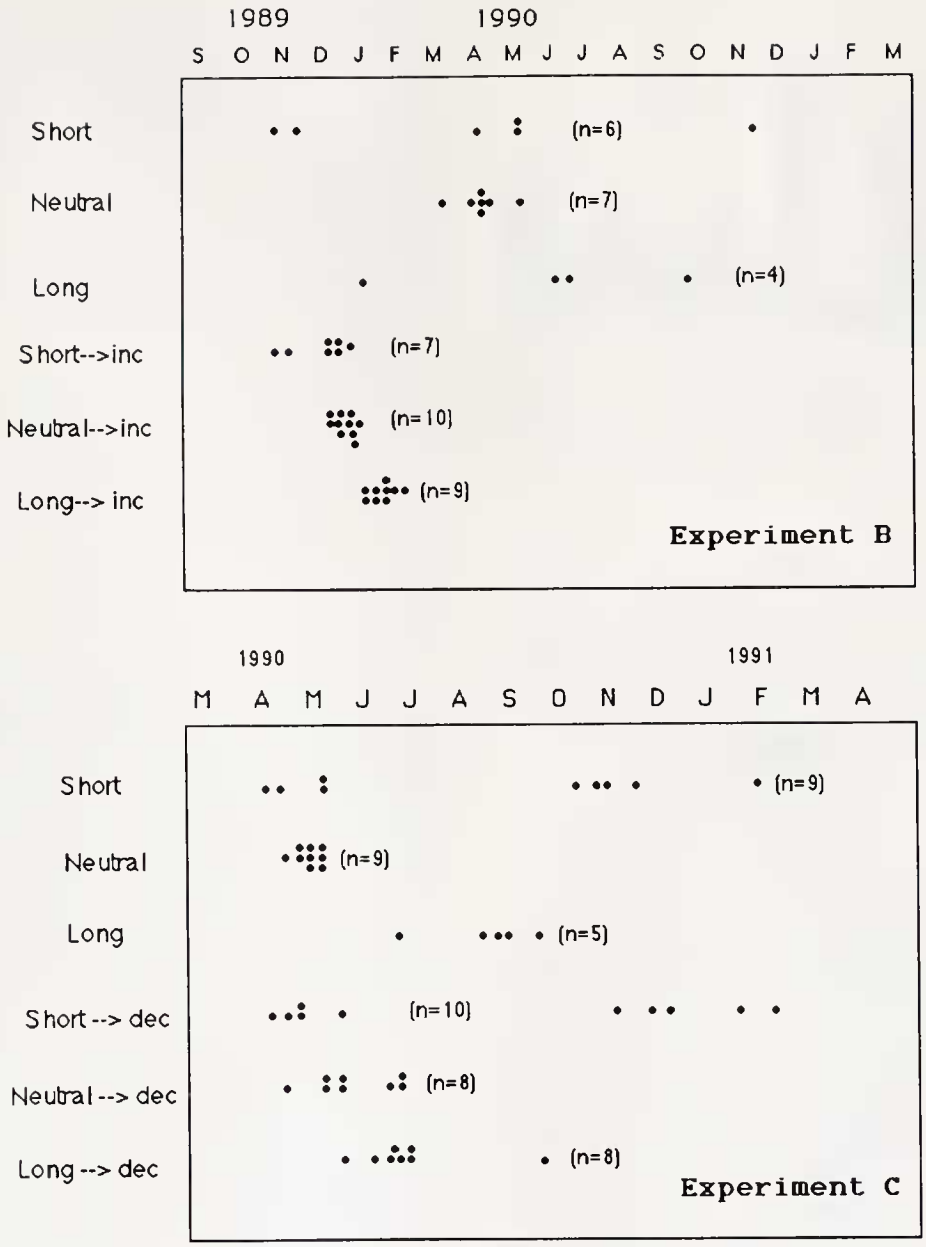


**Figure 3.** Parturition dates of *Neanthes limnicola* exposed to constant short (L:D 8:16), neutral (12:12), or long (16:8) daylength, initial fixed daylength followed by increasing daylength (upper: experiment B), and initial fixed daylength followed by decreasing daylength (lower: experiment C). Worms exposed to increasing daylengths were 6 months out of phase with ambient daylength, thus parturition dates in October, November, December, and January were actually in light regimes corresponding to April, May, June, and July.

Three months of fixed daylength followed by increasing daylength had a significant effect on life span (Table I; one-way ANOVA, $F_{5,37} = 7.73$, $P = 0.0001$). Worms that saw increasing daylength spent the shortest time in culture, and all three pair-wise comparisons of fixed (control) versus increasing daylengths (*e.g.*, fixed short daylength versus short → increasing daylength) showed significant differences (Fisher's LSD, $P < 0.05$ for all three comparisons).

Increasing daylength had a significant effect on fecundity (one-way ANOVA, $F_{5,37} = 7.79$, $P = 0.05$), yet there was no consistent trend (Table I). The highest mean fecundity recorded was in cultures that experienced long → increasing daylengths ($\bar{x} = 152.77$), even though these worms lived from 77 to 132 days less than all three of the fixed-daylength treatments (Fig. 4). High mean fecundity ($\bar{x} = 143.4$) also was recorded in constant neutral

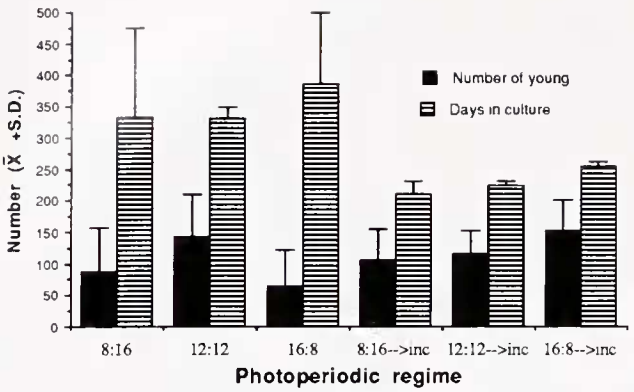


**Figure 4.** Fecundity (number of young born) and life span (days in culture) in experimental light regimes of experiment B. inc: increasing daylengths.

daylength, but the lowest mean fecundities were in constant long ($\bar{x}$ = 64.75) and constant short ($\bar{x}$ = 87.16) daylengths.

Comparison of pooled constant daylength (constant short, neutral, and long) with pooled increasing daylength showed a significant difference in life span. Worms that saw increasing daylengths independent of their initial fixed daylength, reproduced at a younger age than those in constant daylength ($\bar{x}_{constant}$ = 344 days, n = 17, $\bar{x}_{increasing}$ = 230 days, n = 26; t = 5.83, df = 41, $P$ = 0.0001).

*Decreasing daylength experiment (experiment C)*

Seven weeks of fixed daylength followed by two months of decreasing daylength, and then constant short daylength had a significant effect on the timing of parturition (Table I; one-way ANOVA of mean parturition date, $F_{5,43}$ = 2.83, $P$ = 0.02). Cultures in neutral and neutral → decreasing daylength reproduced in the ambient spring and showed much tighter reproductive synchrony than cultures in other light treatments, which reproduced later in the year, on average, and with much greater spread in the parturition dates (Fig. 3).

Life span was affected by decreasing daylengths (Table I; one-way ANOVA, $F_{5,43}$ = 2.76, $P$ = 0.03). Those worms in cultures maintained at neutral and neutral → decreasing daylengths took the shortest time to reproduce ($\bar{x}_{neutral}$ = 265 days, $\bar{x}_{neutral \rightarrow decreasing}$ = 288 days).

Decreasing daylengths affected fecundity (Table I; one-way ANOVA, $F_{5,43}$ = 2.50, $P$ = 0.04). Highest fecundities were recorded in cultures at neutral ($\bar{x}$ = 108.9 young) and neutral → decreasing ($\bar{x}$ = 106.3 young), even though on average, worms in both these treatments had a shorter life span than worms in cultures in other light treatments. Worms in long daylength had the longest life span, but produced the fewest young ($\bar{x}$ = 39.4).

Initial fixed daylength treatments, independent of shifting, had a significant effect on all three parameters. Mean parturition dates are significantly different (one-way ANOVA, $F_{2,46}$ = 5.99, $P$ = 0.004); worms initially exposed to neutral daylengths had a mean parturition date in spring ($\bar{x}$ = 8 May) whereas those initially exposed to short daylength gave birth in spring and fall ($\bar{x}_{short}$ = 6 August) and those in long daylength reproduced in the summer and fall ($\bar{x}_{long}$ = 13 July) (Fig. 3; Fisher's LSD, $P < 0.05$ for both comparisons). Correspondingly, worms exposed to neutral daylength initially have shorter life spans than those exposed initially to either short or long daylength ($\bar{x}_{neutral}$ = 276 days, $\bar{x}_{short}$ = 365 days, $\bar{x}_{long}$ = 348 days; one-way ANOVA, $F_{2,46}$ = 5.92, $P$ = 0.005; Fisher's LSD, $P < 0.05$ for both comparisons). Fecundity was also significantly affected (one-way ANOVA, $F_{2,46}$ = 4.77; $P$ = 0.01); worms initially exposed to neutral daylength produced significantly more young ($\bar{x}$ = 107.64) than worms initially exposed to long ($\bar{x}$ = 63.07; Fisher's LSD, $P < 0.05$), but not to short ($\bar{x}$ = 89.21) daylengths.

## Discussion

We have shown that seasonally changing photoperiod controls the timing of parturition in *Neanthes limnicola* from central California (Fong and Pearse, 1992). In the field, worms give birth mainly in late winter–spring (late February–May), and in the laboratory, parturition can be shifted to late summer–fall when the worms had been reared under seasonally changing photoperiods 6 months out of phase with ambient. In California, winter–spring light regimes increase from about (9.5:14.5 L:D) on 21 December to about (14.5:9.5 L:D) on 21 June. Thus, most worms experience increasing daylengths for 2–5 months before giving birth.

In the present study, worms exposed to increasing daylengths (corresponding to changes in light regimes from February to June) after 3 months of either fixed short, neutral, or long daylengths gave birth within 3–5 months, independent of the initial fixed daylengths to which they were exposed. That increasing daylengths act to synchronize parturitions in *N. limnicola* corresponds to our earlier findings (Fong and Pearse, 1992).

Parturition also was synchronized when the worms were exposed to fixed, neutral (12:12) daylength: nearly all gave birth at 9–11 months of age in all three experiments. In experiment A, most worms in neutral daylength gave birth at 8–10 months of age, but two worms gave birth 10–12 months later in the following late winter–spring. The latter two worms may have missed the "gate-open period," which specifies a time interval in which worms may initiate a rapid phase of oocyte growth (Olive, 1984), and had to wait another full cycle for it to reopen (see below). However, these worms never saw any changes in photoperiod with which to gauge time and synchronize reproduction. Laboratory temperatures did vary, but with little

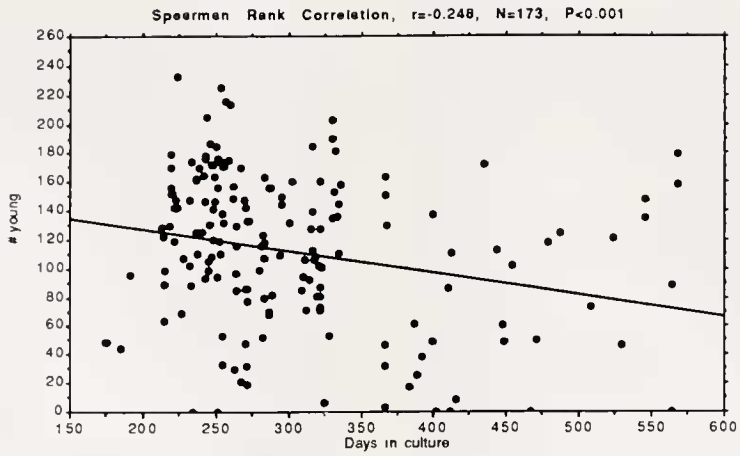


**Figure 5.** Regression of number of young on days in culture. Data are combined from all three experiments.

pattern by which the animals could seasonally synchronize activities (Fig. 1). Parturition by most worms under fixed, neutral daylength was in April–June 1989 in experiment A, following increasing temperatures, and in April–May 1990 in experiments B and C, following a period of little temperature change. Previous experiments with *N. limnicola* (Fong and Pearse, 1992) showed that temperature had no effect on the timing of parturition in worms exposed to seasonally changing daylength from birth.

Constant short (8:16) and long (16:8) daylength had no consistent effect on parturition synchrony but tended to desynchronize parturition especially when long daylength was combined (either before or after) with neutral daylength. Short and long daylengths also had a detrimental effect on fecundity. However, the disruptive effect of long daylength on reproductive timing and fecundity was mitigated when followed by increasing daylengths.

Decreasing daylengths did not synchronize parturition and no consistent trend was evident. Moreover, parturient synchronization was not as tight among worms in neutral → decreasing daylengths as it was in constant neutral daylengths (Fig. 3).

Exposure to different photoperiodic regimes affected fecundity. The highest fecundity in any experimental treatment was found in fixed neutral daylength (experiment A), and the lowest in long daylength (experiment C). Nevertheless, fecundities in the present experiments, in general, were lower than those previously recorded in *N. limnicola* exposed to seasonally changing daylengths continuously from birth (Fong and Pearse, 1992). Thus, a seasonal cycle of decreasing and increasing daylengths may be necessary for maximum fecundity.

Fecundity is a component of fitness. The finding that photoperiod can strongly influence fecundity and hence fitness has also been shown by Chu and Levin (1989) in the spionid polychaete *Streblospio benedicti*. In the case of *N. limnicola*, parturition during periods of increasing daylength appears to select for higher fecundity.

Although both fecundity and life span of *Neanthes limnicola* varied with photoperiod, life span varied inversely with fecundity (Fig. 5). Worms that saw short → increasing and long → increasing daylength (experiment B) had a shorter life span in culture but higher fecundity than the constant short- and constant long-daylength controls (Fig. 4). Likewise in experiment C, worms in neutral and neutral → decreasing daylengths had the highest fecundity but the shortest life span. The worm that gave birth to the most young (232) in all of our experiments lived 224 days, whereas two of the longest-lived worms (564 days each) produced only 88 and 0 young (Fig. 5). These results are inconsistent with life history theory that life span, growth, and fecundity are positively correlated (*e.g.*, Bell, 1980), but consistent with our previous findings (Fong and Pearse, 1992), and indicate that *N. limnicola* can reach maximum reproductive potential in half its lifetime. Consequently, these animals spend the later part of their lives in maintenance, waiting for a "gate-open period," before proceeding with reproduction. This conclusion indicates that the photoperiodic control resulting in seasonality of parturition in spring-early summer is under strong selection.

Our experiments reported here demonstrate that in *Neanthes limnicola* (1) parturition occurs with near-maximum fecundity in about 8–11 months under fixed, neutral photoperiod, (2) parturition is delayed and asynchronous, and fecundity is lowered under short, long, or decreasing photoperiods, and (3) parturition is earlier, synchronous, and only slightly below maximum fecundity when given seasonally increasing photoperiods after an initial 3-month exposure to either short, neutral, or long photoperiod. Moreover, changes in temperature cannot account for the synchrony displayed by the animals held

under neutral photoperiod (see above). These results from the neutral photoperiod treatments suggest that the animals are programmed to complete their life cycle, from birth to metamorphosis and parturition, in late winter and spring. The slight extension (up to 4 months) of the annual rhythm over the underlying 8–11 month endogenous rhythm is probably the result of modifying effects of seasonally changing photoperiod. This situation is similar to that described by Olive and Garwood (1983) for *Nereis diversicolor* in northern England. Worms maintained under constant temperatures and daylengths become sexually mature at the same time as worms in the field. Oocyte growth in *N. diversicolor* proceeds at the same rate at 5°, 10°, 15°, or 20°C, thus the duration of oogenesis is fixed, and the timing of its completion depends on the time of its initiation. At 5° and 10°C, two cycles of reproductive activity occur at intervals somewhat less than 1 year apart. At 15°C, all worms become sexually mature within 1 year of collection. This pattern of reproductive activity suggests an endogenous, gated reproductive rhythm of circannual periodicity, initiated at birth, which free-runs for 1–3 years (Olive and Garwood, 1983; Olive, 1984). No evidence for an exogenous, entraining zeitgeber has yet been found, however.

Carpet beetles (*Anthrenus verbasci*) appear to have a similar endogenous, circannual rhythm of pupation. Beetle larvae maintained in constant darkness at either 22.5° or 25°C, show one pulse of pupation following their first winter diapause, then emerge the following spring. But, larvae held at either 17.5° or 20°C have two peaks of pupation separated by about 41 weeks (Blake, 1959).

The main component of photoperiod that seems important for maintaining reproductive synchrony in populations of *Neanthes limnicola* is increasing daylength, as is normally experienced in the winter and spring. Not only did increasing daylength synchronize reproduction in our experiments reported here, but earlier experiments showed that the life cycle of *N. limnicola* could be shifted out of phase when the animals were held in seasonally changing photoperiods out of phase with ambient (Fong and Pearse, 1992). Similar experiments have shown that both a fall reproductive diapause in the shrimp *Heptacarpus pictus* (Custer, 1986) and gametogenesis in the sea star *Pisaster ochraceus* (Pearse and Eernisse, 1982; Pearse *et al.*, 1986) can be shifted by shifting the phase of the seasonally changing photoperiods, but these reproductive cycles remain unaffected when the animals are maintained under fixed long, neutral, or short daylengths. From the experiments with *P. ochraceus,* Pearse *et al.* (1986) suggested that an underlying endogenous rhythm was synchronized by changing photoperiod. However, unlike *N. limnicola* and *H. pictus,* which live only 1–2 years, individuals of *P. ochraceus* live decades or more, spawning year after year; photoperiodism maintains synchrony among individuals of *P. ochraceus* over many years, while it maintains synchrony within successive generations of *N. limnicola* and *H. pictus.*

In most examples of photoperiodism, including those of seasonal reproductive activity, one or more "critical daylengths" appear to trigger events leading to synchronization (Saunders, 1982). The sea urchin *Strongylocentrotus purpuratus,* a marine example, is gametogenic and full of gametes when under photoperiods less than 12 h, but gametogenesis is repressed under longer daylengths (Bay-Schmith and Pearse, 1987). Thus, gametogenesis is initiated in the fall when daylength drops below 12 h and slows down in the spring when daylength exceeds 12 h. Such examples are the basis for "hour-glass," "circadian," or similar models explaining photoperiodism (Saunders, 1982; Gwinner, 1986); critical processes require a minimum amount of light each day (hour-glass), or a particular length of time between two lighted periods (circadian), to activate a photoperiodic response. However, these models are inadequate for explaining how changing daylengths, but not fixed daylengths of any length [(or expected combinations such as short → long (experiment A)], might synchronize activities such as parturition in *Neanthes limnicola,* diapause in *Heptacarpus pictus,* or gametogenesis in *Pisaster ochraceus* (or antler shedding in reindeer, Goss, 1982, 1984). Rather, the organisms need to be able to measure daylength and compare it with earlier daylengths before initiating a photoperiodic response. As pointed out by Pearse *et al.* (1986), new models and insights are needed to explain how changing daylengths can act to synchronize seasonal activities with an underlying circannual endogenous rhythm.

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